FY 2004 Investigational Report:

Health and Physiological Assessment of VAMP Release Groups – 2004

Rick Harmon, Ken Nichols and J. Scott Foott*
US Fish and Wildlife Service, California-Nevada Fish Health Center, Anderson, CA

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* Direct correspondence:
CA-NV Fish Health Center
24411 Coleman Fish Hatchery Rd
Anderson, CA 96007
(530) 365-4271 Fax: (530) 365-7150

Summary: Juvenile Chinook salmon were sampled directly from the transport truck upon their arrival at Durham Ferry, Mossdale, and Jersey Point release sites (4/22 – 26/04). The incidence of *Tetracapsula bryosalmonae* infection (causative agent of Proliferative Kidney Disease) increased with each successive release group. These infections were rated as moderate and appeared to be early in the disease cycle. Mean plasma cortisol concentration increased with transport time and salmon sampled at Jersey Point had significantly higher values than those at Durham Ferry. Plasma chloride and protein concentrations were not correlated with either *T. bryosalmonae* infection or plasma cortisol levels. Gill sodium-potassium-adenosine triphosphatase activity in Mossdale fish were higher than those at Durham Ferry however a large number of each group had values indicative of parr. Overall health and physiological status of the three release groups was deemed adequate for out-migration evaluation.

Methods

Merced River Hatchery (MRH) Chinook salmon smolts from the Vernalis Adaptive Management Plan (VAMP) study releases were examined for general health and physiological indicators of smolt development, stress, and plasma osmolarity. A total of 96 fish were examined from the 3 release groups following their transport to release sites at Durham Ferry (4/22/04), Mossdale (4/23/04) and Jersey Point (4/26/04). Thirty two salmon were collected from the transport truck upon its arrival at the release site. The first 12 fish were rapidly euthanized with MS222, bled for plasma (chloride, protein, and cortisol), and sampled for kidney and gill tissue. Anterior kidney was used for viral assay, culture of systemic bacteria, and imprint smears later examined for *Renibacterium salmoninarum* by a direct fluorescent antibody test. Posterior kidney was processed for histological sections to evaluate *Tetracapsula bryosalmonae* infection and kidney inflammation. Gill tissue was tested for sodium-potassium-adenosine triphosphatase activity (ATPase). The remaining 20 fish were than sampled for only kidney tissue to perform the above microbiological and histological assays. Both internal and external abnormalities were recorded for each smolt.

Results and discussion

No viral pathogens, cultured systemic bacteria or *R. salmoninarum* were detected in the 96 fish sampled from all three sites. *Tetracapsula bryosalmonae* was detected in 37 % of salmon sampled at Durham Ferry, 50% at Mossdale, and 64% at Jersey Point (Table 1.). Only 14 % or less of the infected kidneys were rated as showing moderate inflammatory changes indicating the population was in an early stage of the disease. Proliferative kidney disease has been observed in MRH Chinook smolts for many years, and the incidence in VAMP release groups has ranged from 4% to 100% in the last 5 years (Table 2). This progressive disease can reduce a fish's performance due to the associated kidney dysfunction and anemia

Table 1. Prevalence of *Tetracapsula bryosalmonae* infection (POI) and kidney inflammation rating (moderate or severe) in 2004 VAMP release groups.

Site	Moderate	Severe	POI
Durham Ferry	5/22 (23)	3/22 (14)	8/22 (37)
Mossdale	10/22 (45)	1/22 (5)	11/22 (50)
Jersey Point	12/22 (55)	2/22 (9)	14/22 (64)

Table 2. Prevalence of *Tetracapsula bryosalmonae* detected in Merced River Hatchery Chinook Salmon smolts 1996-2004. All samples were taken from VAMP (and precursor project) release groups. Fish were assayed by histopathological examination of posterior kidney.

Year	Sample Date(s)	Prevalence
1996	5/01	5/8 (63%)
1997	5/01	0/10 (0%)
1998	4/17	0/6 (0%)
1999	4/20	0/6 (0%)
2000	4/18 – 5/02	2/45 (4%)
2001	5/01 – 5/12	34/34 (100%)
2002	4/19 – 5/01	92/201 (46%)
2003	4/21 – 5/02	30/48 (63%)
2004	4/22 – 4/26	33/66 (50%)

Gill ATPase activity was significantly higher in Durham Ferry fish compared with the Mossdale samples (t-test: t = -2.661 with 22 degrees of freedom, P=0.014). A large percentage of each group (42% and 83%) had ATPase activities associated with parr ($\leq$ 6.7 μmole ADP / mg protein / hr). This data indicates that these fish were not in an advance state of smoltification at the time of release. The effect of this observation on migration behavior is uncertain as gill ATPase activity can change rapidly during out-migration. All groups had similar sized fish (mean fork length 84 – 86 mm).

Plasma cortisol tended to increase with each successive release group (Table 3). Jersey Point fish had significantly higher levels than the Durham Ferry group (ANOVA, P= 0.014). Longer transport time was a likely factor in the observed trend. Plasma protein and chloride values were normal and similar among all groups (Table 3). These two

indicators of osmolarity status were not affected by the early stage of PKD observed in the sampled fish or their cortisol concentration (t-test, t=0.334, 34 d.f., p=0.740).

Table 3. Blood plasma chemistry values for VAMP 2004 release groups sampled at the release site. Smolts were sampled immediately after transport. Data is presented as Mean (standard deviation) with statistical significance denoted by different letters.

Mean(Std) N=12	Durham Ferry 4/22/04	Mossdale 4/23/04	Jersey Point 4/26/04
ATPase	5.6 (1.6) a	7.6 (2.0) ab	N/A
Cortisol	74.3 (47.4) a	109.3 (31.6) ab	136.4 (62.3) b
Chloride	72.9 (5.8)	74.0 (9.6)	74.3 (5.8)
Protein	2.4 (0.9)	2.9 (0.3)	2.9 (0.6)

N/A not available as sample was lost

In summary, the examined salmon were relatively healthy and should perform adequately for an assessment of out-migration survival. Longer transport distances resulted in higher plasma cortisol responses however we did not detect physiological impairment associated with these elevated cortisol levels.

Methods

Chloride determined by colorimetric method using Raichem kit #85133 (San Diego, CA)
Total Protein determined by biuret colorimetric method using Sigma Diagnostics #541-2 (St. Louis, MO)
Cortisol determined by ELISA kit produced by Neogen Corporation (#402710, Lexington, KY)
Na-K-ATPase assay (McCormick, S.D. and H.A. Bern. 1989. In vitro stimulation of Na+-K+-ATPase activity and ouabain binding by cortisol in coho salmon gill. Am. J. Physiol. 256: R707-R715.)